Rejection Under 35 U.S.C. § 112, First Paragraph

Claims 89-100, 109-116, 140, 148-160 and 169-190 stand rejected under 35 U.S.C. § 112, first paragraph. The Office Action acknowledges that Applicants have demonstrated successful *in vitro* gene transfer capabilities in human hematopoietic cells, but questions whether the *in vitro* data can be extrapolated to the *in vivo* environment and to therapeutic applications. In support of this position, it is stated in the Office Action that gene therapy is still in its infancy and is highly unpredictable, and that numerous factors complicate the gene therapy art which have not been shown to overcome by routine experimentation. In view of this, the Office Action concludes, one of skill in the art could not make and use the invention with out undue experimentation.

Applicants respectfully disagree. Applicants assert that one of skill in the art would know how to make and use the claimed invention without undue experimentation. Applicants do not claim gene therapy generally, and cannot speak to the state of gene therapy art generally. Applicants do assert, however, that any such generalization does not apply to the claimed subject matter in view of the significant advances made in gene therapy using cochleate technology, and also in view of successful gene therapy protocols that can be used to extrapolate the *in vitro* results to the *in vivo* environment.

Applicants submit that gene therapy has been successfully employed with respect to polynucleotide-cochleates. For example, Applicants and their collaborators have successfully employed DNA-cochleate vaccines (the DNA encoding the surface glycoprotein from HIV-1), with Sendai virus glycoproteins incorporated into the cochleate lipid bilayers, to affect a strong, long-lasting *in vivo* immune response in mice. See e.g., Gould-Fogerite *et al.*, "Targeting Immune Response Induction With Cochleate and Liposome-Based Vaccines," Advanced Drug Delivery Reviews 32: 273-87 (1998) (IDS Ref. #10). DNA-cochleates without surface glycoprotein also have been employed to mediate strong cellular immune responses. Mannino & Gould-Fogerite, "Antigen Cochleate Preparations for Oral and Systemic Vaccination," NEW GENERATION VACCINES, Chapter 18, pp. 229-237 (Marcel Dekker 2d Ed. 1997)(IDS Ref. #B7)

Indeed, substantial research by Applicants and their collaborators have demonstrated that cochleate formulations are simple, safe, and highly efficacious mediators of the *in vivo* delivery of proteins, peptides, antisense oligonucleotides, and

DNA for the induction of antigen-specific immune responses following oral, intranasal, and intramuscular administration. See e.g., Gould-Fogerite and Mannino, "Cochleates for Induction of Mucosal and Systemic Immune Responses," METHODS IN MOLECULAR MEDICINE Vol. 42, Chapter 10, pp. 179-197 (1999) (IDS Ref. #B3); Mannino & Gould-Fogerite, "Antigen Cochleate Preparations for Oral and Systemic Vaccination," NEW GENERATION VACCINES, Chapter 18, pp. 229-237 (Marcel Dekker 2d Ed. 1997)(IDS Ref. #B7); Michalek *et al.*, "Antigen Delivery Systems: Nonliving Microparticles, Liposomes, Cochleates and ISCOMS," MUCOSAL IMMUNOLOGY, Chapter 47, pp. 759-778 (Academic Press 1999) (IDS Ref. #B8); Parker *et al.*, "*In Vivo* and *In Vitro* Antiprolferative Effects of Antisense Interleukin 10 Oligonucleotides," Methods in Enzymology 313: 411-429 (1999)(IDS Ref. #B9).

Moreover, research by others can readily be used to extrapolate Applicants' *in vitro* results to *ex vivo* and *in vivo* results. *In vitro* manipulation of hematopoietic stem cells, particularly CD34+ cells, is well known to those of ordinary skill in the art. Further, the capability and success of using such cells to achieve gene therapy in animals and humans is well documented. See e.g., Brenner *et al.*, "Gene marking and autologous bone marrow transplantation," ANN N Y ACAD SCI. 716:204-15, 225-7 (May 31, 1994) (IDS Ref. #B1); Kiem *et al.*, "Gene therapy and bone marrow transplantation," CURR OPIN ONCOL. 7(2):107-14 (March 1995) (IDS Ref. #B5).

The ability to manipulate hematopoietic stem cells, taken from an animal or human patient, and the affect of autologously transplanting these manipulated cells has been widely practiced and reported. See e.g., Cavazana-Calvo *et al.*, "Gene Therapy Of Human Severe Combined Immunodeficiency (SCID)=X1 Disease," Science 288(5466):669-672 (2000) (Describing an *ex vivo* autologously transplanted use for infant bone marrow CD34+ cells, which were transduced with a retrovirus-derived vector containing the gene encoding the gamma cytokine receptor subunit of various interleukin receptors; Gamma transgenes and appropriate antigenic-specific responses were detected in two patients demonstrating a clinical benefit for this type of gene therapy) (IDS Ref. #B12); Schiedlmeier *et al.*, "Quantitative Assessment of Retroviral Transfer of the Human Multidrug Resistance 1 Gene to Human Mobilized Peripheral Blood Progenitor Cells Engrafted In Nonobese Diabetic/Severe Combined Immunodeficient Mice," Blood, 95(4):1237-1248 (2000) (Demonstrating successful *ex vivo* gene therapy using human CD34+ cells, transduced with retroviral-mediated cytostatice drug-resistance genes, to

repopulate mice) (IDS Ref. #B11); Parkman *et al.*, "Gene Therapy For Adenosine Deaminase Deficiency," Annual Rev. Med. 51:33-57 (2000) (Clinical gene therapy trials for adenosine deaminase deficiency using hematopoietic stem cells taken from either umbilical cord blood or neonatal bone marrow and using mature T lymphocytes) (IDS Ref. #B10); Inser *et al.*, "Angiogenesis And Vasculogenesis As Therapeutic Strategies For Postnatal Neovascularization," J. Clin. Invest 103(9):1231-1236 (1999) (Demonstrating that CD34+ cells can appropriately populate and differentiate into endothelial cells when transplanted *ex vivo*) (IDS Ref. #B4). Accordingly, Applicants assert that the transduced CD34+ cells disclosed it the instant specification can reasonably be expected to repopulate an animal or human patient.

Finally, Applicants submit that the use of cochleate delivery vehicles does not introduce uncertainty with respect to safety and efficacy in gene therapy applications, and does not give rise to an expectation that Applicants' successful *in vitro* results cannot be extrapolated to gene therapy. Cochleate delivery vehicles are prepared from safe, non-toxic, non-inflammatory, naturally occurring materials. In fact, clinical studies by other investigators to evaluate the potential of lipids, such as phosphatidylserine, as a nutrient supplement demonstrate their safety and indicate that they may play a role in the support of mental functions in the aging brain. See e.g., Lombardi, "Pharmacological treatment with phosphatidyl serine of 40 ambulatory patients with senile dementia syndrome." Minerva Med 80:599-602 (1989) (IDS Ref. #B6).

Significant work also has been devoted to demonstrating the safe and successful delivery of cochleate contents *in vivo*. Cochleate mediated drug delivery in animals by oral and parenteral routes has been demonstrated to be safe and effective at curing animal models of systemic fungal infections (Candidiasis, Aspergillosis, and Cryptococcal meningitis). See e.g., Zarif *et al.*, "Antifungal Activity of Amphotericin B Cochleates Against *Candida albicans* in a Mouse Model," Antimicrobials Agents and Chemotherapy 44:6 1463-1469, 2000 (IDS Ref. #B13); Delmas *et al.*, "Efficacy of orally delivered cochleates containing amphotericin B in a murine model of aspergillosis," Antimicrob Agents Chemother 46(8):2704-7 (2002) (IDS Ref. #B2).

In summary, Applicants submit that studies demonstrating the successful *in vivo* delivery of biologically active molecules in cochleates, and use of CD34+ cells *in vivo* to achieve gene therapy, reasonably allows the extrapolation of Applicants' *in vitro* data to

gene therapy applications. Accordingly, Applicants respectfully submit the rejected claims are enabled and request the withdrawal of the outstanding rejection.

Rejection Under 35 U.S.C. § 103

The Office Action rejects claims 89-99, 109, 111-115, 117-127, 137, 140-145, 148-159, 169, 172-176, 182-184, and 189 stand rejected under 35 U.S.C. § 103(a) as being unpatentable over U.S. Patent No. 6,342,390 to Wiener *et al.* (Weiner) in view of International Patent Publication WO 96 96/25942 to Mannino and Gould-Fogerite (Mannino).

The Office Action acknowledges that Weiner fails to teach a vector delivery structure comprising a cochleate. The outstanding rejection appears to be based on the premise that Mannino teaches cochleates used to transfer DNA and proteins into cells, and that since both Mannino and the primary reference contemplated gene therapy methods, that it would have been obvious to modify Weiner to include the cochleate structure of Mannino. The Office Action reasons that the advantages of cochleates as vehicles for delivery of biologically active molecules taught in Mannino "provides strong motivation to combine the teachings," and that the skilled artisan would have a reasonable expectation of success "in view of the versatility of cochleates as vehicles for transfer of molecules into cells taught" in Mannino.

Applicants respectfully disagree. The disclosure of the advantages of cochleates provided in Mannino provides neither the motivation to modify the liposomes of Weiner, nor a reasonable expectation of success, particularly in view of the marked differences between the liposomes of Weiner and the cochleates of Mannino.

> Where claimed subject matter has been rejected as obvious in view of a combination of prior art references, a proper analysis under §103 requires, *inter alia*, consideration of two factors: (1) whether the prior art would have suggested to those of ordinary skill in the art that they should make the claimed composition or device, or carry out the claimed process; and (2) whether the prior art would also have revealed that in so making or carrying out, those of ordinary skill would have a reasonable expectation of success. Both the suggestion and the reasonable expectation of success must be founded in the prior art, not in the applicant's disclosure.
> In re Vaeck, 947 F.2d 488, 493 (Fed. Cir. 1991).

In re Vaeck (cited in the MPEP, *e.g.*, §§ 2142-2143), concerned an application having claims generally directed to providing for the production of the insecticidal

Bacillus proteins within host cyanobacteria. As summarized in this Federal Circuit opinion, the claims:

> were rejected as unpatentable under *35 U.S.C. § 103* based upon Dzelzkalns in view of Sekar I or Sekar II and Ganesan. The examiner stated that Dzelzkalns discloses a chimeric gene capable of being highly expressed in a cyanobacterium, said gene comprising a promoter region effective for expression in a cyanobacterium operably linked to a structural gene encoding CAT. The examiner acknowledged that the chimeric gene and transformed host of Dzelzkalns differ from the claimed invention in that the former's structural gene encodes CAT rather than insecticidally active protein. However, the examiner pointed out, Sekar I, Sekar II, and Ganesan teach genes encoding insecticidally active proteins produced by *Bacillus*, and the advantages of expressing such genes in heterologous hosts to obtain larger quantities of the protein. The examiner contended that it would have been obvious to one of ordinary skill in the art to substitute the *Bacillus* genes taught by Sekar I, Sekar II, and Ganesan for the CAT gene in the vectors of Dzelzkalns in order to obtain high level expression of the *Bacillus* genes in the transformed cyanobacteria. The examiner further contended that it would have been obvious to use cyanobacteria as heterologous hosts for expression of the claimed genes due to the ability of cyanobacteria to serve as transformed hosts for the expression of heterologous genes. In the absence of evidence to the contrary, the examiner contended, the invention as a whole was prima facie obvious.
> In re Vaeck, 947 F.2d at 491.

The Court found that the U.S. Patent Office (USPTO) had not established the prima facie obviousness of the claims because the cited prior art "simply does not disclose or suggest the expression in cyanobacteria of a chimeric gene encoding an insecticidally active protein, or convey to those of ordinary skill a reasonable expectation of success in doing so." In re Vaeck, 947 F.2d at 493. According to the opinion, the USPTO had attempted to remedy the lack of disclosure or suggestion of the modification by emphasizing the similarity between bacteria and cyanobacteria and argued that this similarity would suggest the use of cyanobacteria as a host for the claimed genes. The Court found that bacteria and cyanobacteria were not viewed as interchangeable by those in the art, and that in any event, similarity alone was not sufficient to motivate the modification. In re Vaeck, 947 F.2d at 493.

Similar to In re Vaeck, there is no disclosure or suggestion in either cited reference to modify Weiner to realize the claimed invention. Contrary to the assertion made in the outstanding Office Action, the recitation of advantages of cochleates as vehicles for delivery of biologically active materials in Mannino provides neither the suggestion nor the motivation to combine the references required to establish

obviousness. The mere fact that Mannino and Weiner can be combined or modified does not render the claimed combination obvious unless the references also suggest the desirability of the combination. MPEP 2143.01, *citing, In re Mills*, 916 F.2d 680 (Fed. Cir. 1990).

Also similar to the situation in In re Vaeck (where bacteria and cyanobacteria were not viewed as interchangeable), cochleates and liposomes are not considered to be interchangeable at least because cochleates and liposomes have fundamentally different structures, properties, and delivery mechanisms. This is true today, and was even more true during the relevant time period, the time of filing, when far less was known about cochleates.

For example, liposomes and cochleates have very different structures and properties. Liposomes in general are a colloidal suspension, and generally are comprised of one or more fluid lipid bilayer membranes surrounding an aqueous interior. In contrast, cochleates are stable lipid–cation precipitates, which are structurally distinct from liposomes. Their unique structure generally includes large, continuous, solid, lipid bilayer sheets stacked and/or rolled up in a spiral configuration, with little or no internal aqueous space. Cations maintain the cochleate in its rolled form, bridging each successive layer, through ionic interaction with opposing bilayers. See e.g., Gould-Fogerite *et al.*, "Targeting Immune Response Induction With Cochleate and Liposome-Based Vaccines," Advanced Drug Delivery Reviews 32: 273-87 (1998) (IDS Ref. #10).

These marked and distinct structural differences cause liposomes and cochleates to interact with their environment in fundamentally different ways. For example, liposomes generally do not spontaneously fuse with other membrane-bound structures (*e.g.*, cells) as do cochleates. Liposome fluid bilayer membranes are highly thermodynamically stable. As such, they resist fusion with other fluid membrane bilayers. Therefore, in general, when an unmodified liposome containing a biologically active agent encounters a target cell membrane, little or no direct transfer of the biologically active agent to the target cell takes place.

In contrast, cochleates deliver their contents to target cells. Cochleates can be envisioned as membrane fusion intermediates. When a cochleate comes into close approximation to a target membrane, a fusion event between the outer layer of the cochleate and the cell membrane occurs. This fusion results in delivery of a small

amount of the encochleated material into the cytoplasm of the target cell. The cochleate may slowly fuse or break free of the cell and be available for another fusion event, either with the same or another cell. See e.g., Mannino & Gould-Fogerite, "Antigen Cochleate Preparations for Oral and Systemic Vaccination," NEW GENERATION VACCINES, Chapter 18, pp. 229-237 (Marcel Dekker 2d Ed. 1997)(IDS Ref. #B7). It also is believed that cochleates can be taken up by endocytosis and fuse from within endocytic vesicles.

Another example is the differences in stability of cochleates as compared to liposomes. The fluid lipid bilayer membrane of liposomes is susceptible to attack from harsh environmental conditions, such as the extremes of pH found in the stomach, and the presence of enzymes and lipases that digest lipid. Once the membrane has been compromised, degradative enzymes (*e.g.*, proteases and nucleases), can attack and destroy the materials within the liposome structure. In contrast, since cochleates generally comprise a series of solid layers, components within the interior of a cochleate structure remain intact, even throughout the outer layers of the cochleate when exposed to harsh environmental conditions or enzymes. Cation concentrations *in vivo* in serum and mucosal secretions are such that the cochleate structure can be maintained.

It was not known prior to Applicants' invention, whether the claimed combination would be successful, largely because the structure, delivery mechanisms and stability of cochleates and liposomes are so different. Specifically, it was not known whether integrative proteins and DNA could be encochleated and delivered intact to a cell: The complexes disclosed in Weiner were encapsulated in and delivered from a liposomal aqueous interior, as opposed to the substantially water free cochleate (where species must exist within the lipid bilayers and/or in the cation bridge between the bilayers).

Based on the foregoing, Applicants respectfully submit that neither the disclosure or suggestion of the claimed invention, nor the reasonable expectation of success can be found in the cited references. As was confirmed in In re Vaeck, "[b]oth the suggestion and the reasonable expectation of success must be founded in the prior art, not in the applicant's disclosure." In re Vaeck 947 at 493.

Accordingly, Applicants submit that the rejected claims are nonobvious at least because the references not only fail to suggest or motivate the claimed subject matter to those of ordinary skill in the art, but also fail to provide a reasonable expectation of

success in making or carrying out the claimed invention. Applicants respectfully request that the obviousness rejection be withdrawn.

Nonstatutory Obviousness-Type Double Patenting Rejection

Claims 89-100 and 109-116 stand rejected under the judicially created doctrine of double patenting over claims 1-28 of U.S. Patent No. 6,340,591. The Office Action indicates that a timely filed terminal disclaimer in compliance with 37 C.F.R. § 1.321(c) may be used to overcome a rejection based on a non-statutory double patenting ground, provided the conflicting application is shown to be commonly owned with this application. Applicants will address the double patenting issue upon a finding that the claimed subject matter is allowable but for the double patenting rejection.

CONCLUSION

In view of the foregoing remarks, reconsideration of the rejections and allowance of all pending claims is respectfully requested. If there are any remaining issues or the Examiner believes that a telephone conversation with Applicants' Attorney would be helpful in expediting prosecution of this application, the Examiner is invited to call the undersigned at (617) 227-7400.

Respectfully submitted,

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